

AMENDMENT

Please amend the subject application as follows:

IN THE CLAIMS:

Please amend the claims as follows:

1. (Currently amended) A method of detecting a target nucleic acid sequence in a sample suspected of comprising the target nucleic acid, the method comprising:

providing a hybridization complex comprising (a) a capture probe that is attached to a solid surface and (b) ~~[[a]] the target nucleic acid sequence that is hybridized to the capture probe, and (c) wherein the target nucleic acid sequence additionally comprises at least one nanoparticle attached to the target nucleic acid sequence, further wherein the nanoparticle is comprises an approximately spherical, metal-atom entity having~~ has a diameter of less than 1000 nanometers, and comprises a material known to absorb light at one or more particular wavelengthsexhibits one of surface plasmon resonance and an interband transition, and wherein the capture probe comprises at least one oligonucleotide ~~sequence that specifically hybridizes~~ is complementary in whole or in part to the target nucleic acid sequence under stringent hybridization conditions;

exposing the solid surface to light at a wavelength absorbed by the nanoparticle and not by the solid surface; and

detecting a temperature of the solid surface in the presence of said complex;

providing a background temperature of the solid surface in the absence of said complex; and

comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature in the presence of said complex relative to ~~[[a]]the background temperature of the solid surface that would be detected in the absence of said complex~~ indicates the presence or amount of target nucleic acid ~~sequence hybridized to the solid surface~~ in the sample.

2. (Currently amended) The method according to claim 1, comprising:
hybridizing ~~[[a]] the target nucleic acid~~sequence to at least one capture probe to form a first hybridization complex, wherein the capture probe is attached to a solid surface at an attachment location;

~~hybridizing~~ attaching a detection probe to the first hybridization complex to form a second hybridization complex, wherein the detection probe comprises ~~[[a]] the~~ nanoparticle;

exposing the solid surface to light at a wavelength absorbed by the nanoparticle; and

detecting the temperature of the solid surface at the attachment location of the capture probe, wherein an increase in temperature at the attachment location as compared to the background temperature of the solid surface indicates hybridization of the target nucleic acid~~sequence to the solid surface~~.

3. (Currently amended) The method of Claim 1, wherein the target nucleic acid~~sequence~~ comprises RNA.

4. (Currently amended) The method of Claim 1, wherein the target nucleic acid~~sequence~~ comprises cDNA.

5. (Original) The method according to Claim 1, wherein the solid surface comprises indium tin oxide.

6. (Currently amended) The method according to Claim 1, wherein the ~~target sequence is present in~~ sample is a biological sample.

7. (Currently amended) The method according to Claim 2, wherein the ~~detection probe comprises a nanoparticle comprises~~ing one or more of the group consisting of a metal~~[[s]] and a metal oxide~~~~[[s]]~~.

8. (Currently amended) The method according to Claim 7, wherein the nanoparticle comprises a metal comprising one or more of the group consisting of gold, silver, and platinum.

9. (Original) The method according to Claim 1, wherein the nanoparticle comprises gold.

10. (Original) The method according to Claim 1, wherein the nanoparticle is a nanoshell.

11. (Currently amended) The method according to Claim 1, wherein the nanoparticle is ~~approximately spherical and~~ has a diameter from about 10 to about 20 nanometers.

12. (Original) The method according to Claim 1, wherein the nanoparticle exhibits surface plasmon resonance, and wherein the solid surface is exposed to light at a wavelength that matches the surface plasmon resonance of the nanoparticle.

13. (Original) The method according to Claim 1, wherein the light is generated by a laser.

14. (Original) The method according to Claim 2, wherein the detection probe further comprises an oligonucleotide attached to the nanoparticle.

15. (Currently amended) The method according to Claim 14, wherein the capture probe is complementary to a first target domain of the target nucleic acid~~sequence~~, and the detection probe oligonucleotide is complementary to a second target domain of the target nucleic acid~~sequence~~.

16. (Currently amended) The method according to Claim 2, wherein the detection probe comprises [[a]] the nanoparticle attached to one partner of a ligand-binding pair, and the target nucleic acid~~sequence~~ comprises the other partner of a ligand-binding pair.

17. (Currently amended) The method according to Claim 16, wherein one partner of [[a]] the ligand-binding pair is streptavidin, and the other partner of the ligand-binding pair is biotin.

18. (Currently amended) The method according to Claim 16, wherein the target nucleic acid ~~sequence~~ comprises biotin.

19. (Currently amended) The method according to claim 18, wherein the biotin has been incorporated into the target nucleic acid~~sequence~~ during nucleic acid amplification.

20. (Original) The method according to Claim 18, wherein the detection probe comprises a nanoparticle attached to streptavidin.

21. (Original) The method according to Claim 1, wherein a plurality of different capture probes are attached to the solid surface in an array, and the location of each capture probe comprises an array element.

22. (Original) The method according to Claim 21, wherein each array element is exposed to light separately.

23. (Original) The method according to Claim 21, wherein the entire plurality of capture probes is exposed to light simultaneously.

24. (Currently amended) The method according to Claim 1, wherein the light is provided by a light source ~~[[is]]~~ selected from the group consisting of a tungsten halogen light source, a xenon arc lamp and a laser.

25. (Original) The method according to Claim 1, where in the exposing is by rastering.

26. (Currently amended) The method according to Claim 1, wherein the target nucleic acid~~sequence~~ is selected from the group consisting of an mRNA ~~sequence derived from a sample~~ and a cDNA ~~sequence derived from a sample~~.

27. (Currently amended) The method according to Claim 1, wherein the capture probe comprises an oligonucleotide ~~sequence~~ from a gene of interest.

28. (Currently amended) The method according to Claim 1, wherein the capture probe comprises ~~or is suspected to comprise~~ a mutation to be detected.

29. (Currently amended) The method according to Claim 1, wherein the target nucleic acid ~~sequence~~ comprises or is suspected to comprise a mutation to be detected.

30. (Original) The method according to Claim 1, wherein the nanoparticle comprises silver and the solid surface is exposed to light at a wavelength ranging from about 420-460 nm.

31. (Original) The method according to Claim 1, wherein the nanoparticle comprises gold and the solid surface is exposed to light at a wavelength of about 532 nm.

32. (Original) The method according to Claim 1, wherein the detecting step is carried out by a thermocouple attached to a side of the solid surface upon which capture probes are not attached.

33. (Original) The method according to Claim 1, wherein the detecting step is carried out by infrared thermography.

34. (Original) The method according to Claim 1, wherein the detecting step is carried out by Fourier Transform infrared thermography.

35. (Original) The method according to Claim 1, wherein the detecting step comprises capturing a thermal image by means of an infrared camera.

36. (Original) The method according to Claim 1, wherein the detecting step is carried out by a charge coupled device.

37. (Canceled)

38. (Currently amended) The method according to claim ~~[[37]]~~1, where the nanoparticle is attached to the target nucleic acid~~sequence~~ by one of the group consisting of a binding pair and complementary nucleic acids.

39. (Currently amended) The method according to claim ~~[[37]]~~1, where the nanoparticle is attached to the target nucleic acid~~sequence~~ by one of the group consisting of primer extension and ligation of a nanoparticle-labeled nucleic acid.

40. (Currently amended) The method of claim 1, wherein the hybridization complex comprises a detection probe, wherein the detection probe comprises the nanoparticle.

41. (Currently amended) The method of claim 40, wherein the detection probe is attached to the target nucleic acid~~sequence~~ before, during, or after the target nucleic acid~~sequence~~ hybridizes to the capture probe.

42. (Currently amended) The method of claim 40, comprising the sequential steps of hybridizing the target nucleic acid to the capture probe to form a hybrid; and then reacting the hybrid with a detection probe.

43. (Previously presented) The method of claim 1, wherein the hybridization complex is present at a concentration of at least 10 fM.

44. (Currently amended) A method of detecting a target nucleic acid sequence in a sample suspected of comprising the target nucleic acid, the method comprising:

providing an at least 10 fM concentration of a hybridization complex comprising (a) a capture probe that is attached to a solid surface and (b) a target nucleic acid ~~sequence that is hybridized to the capture probe, and (c) wherein the target nucleic acid sequence additionally comprises~~ at least one nanoparticle attached to the target nucleic acid ~~sequence~~, further wherein the nanoparticle comprises a metal or metal oxide that exhibits surface plasmon resonance, is ~~approximately spherical~~, and has a diameter of less than 1000 nanometers, the solid surface is a material that is different than the nanoparticle, and wherein the capture probe comprises at least one oligonucleotide sequence that ~~specifically hybridizes~~ is complementary in whole or in part to the target nucleic acid sequence under stringent hybridization conditions;

exposing the solid surface to light at a wavelength that matches the surface plasmon resonance of the nanoparticle and is not absorbed by the solid surface; and

detecting a temperature of the solid surface in the presence of said complex;

providing a background temperature of the solid surface in the absence of said complex; and

comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature of the solid surface in the presence of said complex relative to ~~[[a]] the background temperature of the solid surface that would be detected in the absence of said complex~~ indicates the presence or amount of target nucleic acid sequence hybridized to the solid surface in the sample.

45. (Currently amended) A method of detecting a target nucleic acid sequence in a sample suspected of comprising the target nucleic acid, the method comprising:

providing an at least ~~least~~ 10 fM concentration of a hybridization complex comprising (a) a capture probe that is attached to a solid surface ~~and~~ (b) a target nucleic acid sequence ~~that is hybridized to the capture probe, and (c) wherein the target nucleic acid sequence additionally comprises~~ at least one approximately spherical gold nanoparticle having a diameter of between 5 and 200 nm attached to the target nucleic acid sequence;

exposing the solid surface to light at a wavelength of between about 510 nm and about 560 nm; and

detecting a temperature of the solid surface in the presence of said complex;

providing a background temperature of the solid surface in the absence of said complex; and

comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature of the solid surface in the presence of said complex relative to ~~[[a]] the background temperature of the solid surface that would be detected in the absence of said complex~~ indicates the presence or amount of target nucleic acid sequence hybridized to the solid surface in the sample;

wherein providing a hybridization complex further comprises:

providing a capture probe attached to a solid surface selected from glass and indium tin oxide (ITO), the capture probe comprising an oligonucleotide of 5 to 50 nucleotides ~~sequence~~ that is complementary to the target nucleic acid ~~sequence~~;

providing a target nucleic acid ~~sequence~~ comprising an oligonucleotide ~~sequence~~ of between 10 and 300 nucleotides;

incubating the target nucleic acid ~~sequence~~ with the capture probe at a sodium ion concentration of less than 1.0 M, a pH of between 7.0 and 8.3, and at a temperature of at least 30°C; and

removing any unhybridized ~~target-nucleic acid sequence~~.

Please add the following new claim:

46. (New) The method of claim 1, wherein the sample comprises a heterogeneous nucleic acid mixture and providing the hybridization complex comprises selecting hybridization conditions wherein the capture probe hybridizes to the target nucleic acid to the exclusion of other nucleic acids present in the sample.